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<110> KaloBios, Inc.

<120> TRANSACTIVATION SYSTEM FOR MAMMALIAN CELLS

<130> CELA001/01WO

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<170> PatentIn version 3.2

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Ala Gly Asp Asp Phe Ser Arg Arg Tyr Arg Arg Asp Phe Ala Glu Met 100 \$105\$ 110

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Val Ala Phe Phe Glu Phe Gly Gly Val Met Cys Val Glu Ser Val Asn 145 150 155 160

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Tyr Leu Asn Arg His Leu His Thr Trp Ile Gln Asp Asn Gly Gly Trp
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Gly Val Asn Trp Gly Arg Ile Val Ala Phe Phe Glu Phe Gly Gly Val

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- Ile Phe Pro Asp Ser Val Met Leu Ala Val Gln Glu Gly Ile Asp Leu 65 70 75 80
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- Pro Asn Leu Val Pro Glu Val Ile Asp Leu Thr Gly His Glu Ala Gly 115 120 125
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- Arg Thr Cys Gly Met Phe Val Tyr Ser Pro Val Ser Glu Pro Glu Pro 180 185 190
- Glu Pro Glu Pro Glu Pro Glu Pro Ala Arg Pro Thr Arg Arg Pro Lys 195 200 205
- Met Ala Pro Ala Ile Leu Arg Arg Pro Thr Ser Pro Val Ser Arg Glu 210 215 220
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Phe Glu Glu Thr Glu Glu Pro Asp Phe Thr Ala Leu Cys Gln Lys Leu 50 55 60

Lys Ile Pro Asp His Val Arg Glu Arg Ala Trp Leu Thr Trp Glu Lys 65 70 75 80

Val Ser Ser Val Asp Gly Val Leu Gly Gly Tyr Ile Gln Lys Lys 85 90 95

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cccgaattcg ccgccaccat gaccatggac tctggagcag aca 43

<210> 23

<211> 30

<212> DNA

<213> Artificial

<220>

<223> primer

<400> 23

gtcgacccaa attaatctga tttgtggcag

30

Primer 14: GTCAAGCAAGCTTGCCGCCACCATGAGACATATTATCTGCCACGG (SEQ ID NO: 24)

Primer 15: CGCAGTCTCGAGTTATGGCCTGGGGCGTTTACAGCTC (SEQ ID NO: 25)

Primer 16: CACCTACCCTTCACGAACTGCATGATTTAGACGTGACGGCC (SEQ ID NO: 26)

Primer 17: GGCCGTCACGTCTAAATCATGCAGTTCGTGAAGGGTAGGTG (SEQ ID NO: 27)

Primer 18: CGGAGGTGATCGTTACCGGCCACGAGGCTGGCTTTCCAC (SEQ

ID NO: 28)

Primer 19: GTGGAAAGCCAGCCTCGTGGCCGGTAAGATCGATCACCTCCG (SEQ

ID NO: 29)

Primer 20: GTCAAGCAAGCTTGCCGCCACCATGACCATGGAATCTGGAGC (SEQ

ID NO: 30)

Primer 21: CGCAGTGGATCCTTAATCTGATTTGTGGCAGTAAAGG (SEQ ID NO: 31)

Primer 22: GTCATTCAAAATTTTCCTGAAGGAAGGCCTCCTTGAAAG (SEQ ID

NO: 32)

Primer 23: TCTTTCAAGGAGGCCTTCCTTCAGGAAAATTTTGAATGAC (SEQ ID

NO: 33)

Primer 24: GGCATTCCAAGCTTACTGTTGGTAAAGCCGCCACCATGGAGGCTTGGG

AGTGTTTGG (SEQ ID NO: 34)

Primer 25: GATCGACTCTAGATCATTCCCGAGGGTCCAGGCCGG (SEQ ID NO:

35)

Primer 26: TAAAGCCACCATGGCTCAAGCTGGGAGAACAGGGTATG (SEQ ID

NO: 36)

Primer 27: GATCGACTCTAGATCACTTGTGGCCCAGGTAGGTACCC (SEQ ID

NO: 37)

Primer 28: GTGGGAGATGTGGACGCCGCGGCCGCGCCGCGGCCCCGTGCCAC

CTGTGGTCC (SEQ ID NO: 38)

ATCTCCCAC (SEQ ID NO: 39)

(From Figure 10) Nucleotide coding sequence of Ela cDNA (SEQ

ID NO: 40):

1 aagettgeeg eeaceatgag acatattate tgeeacggag gtgttattae *HindIII* M R H I I C H G G V I

51 cgaagaaatg gccgccagtc ttttggacca gctgatcgaa gaggtactgg T E E M A A S L L D Q L I E E V L

101 ctgataatct tccacctcct agccattttg aaccacctac ccttcacgaa A D N L P P P S H F E P P T L H E

151 ctgtatgatt tagacgtgac ggcccccgaa gatcccaacg aggaggcggt L \underline{Y} D L D V T A P E D P N E E A

201 ttegcagatt ttteccgact etgtaatgtt ggeggtgeag gaagggattg V S Q I F P D S V M L A V Q E G I

251 acttactcac ttttccgccg gcgcccggtt ctccggagcc gcctcacctt D L L T F P P A P G S P E P P H L

- 301 tcccggcagc ccgagcagcc ggagcagaga gccttgggtc cggtttctat S R Q P E Q P E Q R A L G P V S
- 351 gccaaacctt gtaccggagg tgatcgatct tacctgccac gaggctggct MPNLVPEVIDLTCHEAG
- 401 ttccacccag tgacgacgag gatgaagagg gtgaggagtt tgtgttagat FPPSDDEDEEGEFVLD
- 451 tatgtggagc accccgggca cggttgcagg tcttgtcatt atcaccggag Y V E H P G H G C R S C H Y H R
- 501 gaatacgggg gacccagata ttatgtgttc gctttgctat atgaggacct R N T G D P D I M C S L C Y M R T
- 551 gtggcatgtt tgtctacagt cctgtgtctg aacctgagcc tgagcccgag C G M F V Y S P V S E P E P E P E
- 601 ccagaaccgg agcetgeaag acctacccge cgtcctaaaa tggcgcctgc P E P A R P T R R P K M A P
- 651 tatcctgaga cgcccgacat cacctgtgtc tagagaatgc aatagtagta A I L R R P T S P V S R E C N S S
- 701 cggatagetg tgaeteeggt cettetaaca caceteetga gatacaceeg T D S C D S G P S N T P P E I H P
- 751 gtggtcccgc tgtgccccat taaaccagtt gccgtgagag ttggtgggcg V V P L C P I K P V A V R V G G
- 801 tegecagget gtggaatgta tegaggaett gettaaegag eetgggeaae R R Q A V E C I E D L L N E P G Q
- 851 ctttggactt gagctgtaaa cgccccaggc cataactcga g P L D L S C K R P R P - Xhol

(From Figure 11) Nucleotide coding sequence of ElA mutant Y47H (SEQ ID NO: 41):

- 51 cgaagaaatg gccgccagtc ttttggacca gctgatcgaa gaggtactgg T E E M A A S L L D Q L I E E V L
- 101 ctgataatct tccacctcct agccattttg aaccacctac ccttcacgaa A D N L P P P S H F E P P T L H E
- 151 ctgcatgatt tagacgtgac ggccccgaa gatcccaacg aggaggcggt L H D L D V T A P E D P N E E A
- 201 ttcgcagatt tttcccgact ctgtaatgtt ggcggtgcag gaagggattg V S Q I F P D S V M L A V Q E G I
- 251 acttactcac ttttccgccg gcgcccggtt ctccggagcc gcctcacett D L L T F P P A P G S P E P P H L
- 301 tcccggcagc ccgagcagcc ggagcagaga gccttgggtc cggtttctat S R Q P E Q P E Q R A L G P V S
- 351 gccaaacctt gtaccggagg tgatcgatct tacctgccac gaggctggct

MPNL VPE VID LTCH EAG

- 401 ttccacccag tgacgacgag gatgaagagg gtgaggagtt tgtgttagat FPPSDDEDEEGEFVLD
- 451 tatgtggagc accccgggca cggttgcagg tcttgtcatt atcaccggag Y V E H P G H G C R S C H Y H R
- 501 gaatacgggg gacccagata ttatgtgttc gctttgctat atgaggacct R N T G D P D I M C S L C Y M R T
- 551 gtggcatgtt tgtctacagt cctgtgtctg aacctgagcc tgagcccgag C G M F V Y S P V S E P E P E P E
- 601 ccagaaccgg agcetgeaag acctaccge egteetaaaa tggegeetge P E P A R P T R R P K M A P
- 651 tatcctgaga cgcccgacat cacctgtgtc tagagaatgc aatagtagta A I L R R P T S P V S R E C N S S
- 701 cggatagetg tgaeteeggt cettetaaca caceteetga gatacaceeg T D S C D S G P S N T P P E I H P
- 751 gtggtcccgc tgtgccccat taaaccagtt gccgtgagag ttggtgggcg V V P L C P I K P V A V R V G G
- 801 tcgccaggct gtggaatgta tcgaggactt gcttaacgag cctgggcaac R R Q A V E C I E D L L N E P G Q
- 851 ctttggactt gagctgtaaa cgccccaggc cataactcga g P L D L S C K R P R P - XhoI

(From Figure 12) Nucleotide coding sequence of hamster CREB-B cDNA (SEQ ID NO: 42):

- 1 aagettgeeg eeaceatgae eatggaatet ggageagaea aeeageagag HindIII M T M E S G A D N Q Q
- 51 tggagatgct gctgtaacag aagctgaaaa tcaacaaatg acagctcaag S G D A A V T E A E N Q Q M T A Q
- 101 cccaaccaca gattgccaca ttagcccagg tatccatgcc agcagctcat A Q P Q I A T L A Q V S M P A A H
- 151 gegacateat etgeteceae tgtaacetta gtgeagetge eeaatgggea ATSSAPTVTLVQLPNG
- 201 gacagtccaa gtccatggag ttattcaggc ggcccagcca tcagttattc Q T V Q V H G V I Q A A Q P S V I
- 251 agtetecaca agtecaaaca gtteagtett eetgtaagga ettaaaaaga Q S P Q V Q T V Q S S C K D L K R
- 301 cttttctccg gaactcagat ttcaactatt gcagaaagtg aggattcaca L F S G T Q I S T I A E S E D S
- 351 ggaatctgtg gatagtgtaa ctgattccca aaagcgaagg gaaattcttt Q E S V D S V T D S Q K R R E I L
- 401 caaggaggcc ttcctacagg aaaattttga atgacttatc ttctgatgca

SRR PSYR KIL NDL SSDA

- 451 ccaggggtgc caaggattga agaagaaaag tcggaagagg agacttcagc P G V P R I E E E K S E E E T S
- 501 ccctgccatc accactgtga cagtgccaac tccgatttac cagacaagca A P A I T T V T V P T P I Y Q T S
- 551 gtgggcagta tattgccatt acccagggag gagctataca gctggctaac S G Q Y I A I T Q G G A I Q L A N
- 601 aatggtaccg atggggtaca gggccttcag acattaacca tgaccaatgc N G T D G V Q G L Q T L T M T N
- 651 agetgecact cageegggta ceaetattet acagtatgea cagaceactg A A A T Q P G T T I L Q Y A Q T T
- 701 atggacagca gattetagtg eccagcaace aagttgttgt teaagetgee D G Q Q I L V P S N Q V V V Q A A
- 751 tetggegatg tacaaacata ccaaattegt acageaceca etageaceat S G D V Q T Y Q I R T A P T S T
- 801 cgcccctgga gttgttatgg catcctcccc agcacttcct acgcagcctg I A P G V V M A S S P A L P T Q P
- 851 ctgaagaagc agcccggaag agagaggttc gtctaatgaa gaacagggaa A E E A A R K R E V R L M K N R E
- 901 gcagcaagag aatgtcgtag aaagaagaaa gaatatgtga aatgtttaga AARECRRKKEYVKCL
- 951 gaacagagtg gcagtgcttg aaaaccaaaa caagacattg attgaggagc E N R V A V L E N Q N K T L I E E
- 1001 taaaagcact taaggacctt tactgccaca aatcagatta aggatcc L K A L K D L Y C H K S D BamHI

(From Figure 13) Nucleotide coding sequence of hamster CREB-B mutant Y134F (SEQ ID NO: 43):

- 51 tggagatget getgtaacag aagetgaaaa teaacaaatg acageteaag S G D A A V T E A E N Q Q M T A Q
- 101 cccaaccaca gattgccaca ttagcccagg tatccatgcc agcagctcat A Q P Q I A T L A Q V S M P A A H
- 151 gegacateat etgeteceae tgtaacetta gtgeagetge eeaatgggea ATSSAPTVTLVQLPNG
- 201 gacagtccaa gtccatggag ttattcaggc ggcccagcca tcagttattc Q T V Q V H G V I Q A A Q P S V I
- 251 agtetecaca agteca
aaca gtteagtett eetgtaagga ettaaaaaga Q S P Q V Q T V Q S S C K D L K R
- 301 cttttctccg gaactcagat ttcaactatt gcagaaagtg aggattcaca L F S G T Q I S T I A E S E D S

- 351 ggaatctgtg gatagtgtaa ctgattccca aaagcgaagg gaaattcttt Q E S V D S V T D S Q K R R E I L
- 401 caaggaggee tteeteeagg aaaattttga atgaettate ttetgatgea S R R P S F R K I L N D L S S D A
- 451 ccaggggtgc caaggattga agaagaa
aag tcggaagagg agacttcagc P G V P R I E E E K S E E E T S
- 501 ccctgccatc accactgtga cagtgccaac tccgatttac cagacaagca A P A I T T V T V P T P I Y Q T S
- 551 gtgggcagta tattgccatt acccagggag gagctataca gctggctaac S G Q Y I A I T Q G G A I Q L A N
- 601 aatggtaccg atggggtaca gggccttcag acattaacca tgaccaatgc N G T D G V Q G L Q T L T M T N
- 651 agetgecaet cageegggta ceaetattet acagtatgea cagaceaetg AAATQPGTTLLQYAQTT
- 701 atggacagca gattetagtg cccagcaacc aagttgttgt tcaagctgcc D G Q Q I L V P S N Q V V V Q A A
- 751 tetggegatg tacaaacata ccaaattegt acageaceca etageaceat S G D V Q T Y Q I R T A P T S T
- 801 cgcccctgga gttgttatgg catcctcccc agcacttcct acgcagcctg I A P G V V M A S S P A L P T Q P
- 851 ctgaagaagc agcccggaag agagaggttc gtctaatgaa gaacagggaa A E E A A R K R E V R L M K N R E
- 901 gcagcaagag aatgtcgtag aaagaagaaa gaatatgtga aatgtttaga AARECRRKKEYVKCL
- 951 gaacagagtg gcagtgcttg aaaaccaaaa caagacattg attgaggagc ENRVAVLENQNKTLIEE
- 1001 taaaagcact taaggacett tactgccaca aatcagatta aggatee
 L K A L K D L Y C H K S D BamHI

(From Figure 14) Nucleotide coding sequence of E1b-19K (SEQ ID NO: 44):

- 1 <u>aagett</u>actg ttggtaaage egecaceatg gaggettggg agtgtttgga HindIII M E A W E C L
- 51 agatttttct gctgtgcgta acttgctgga acagagctct aacagtacct E D F S A V R N L L E Q S S N S T
- 101 cttggttttg gaggtttctg tggggctcat cccaggcaaa gttagtctgc S W F W R F L W G S S Q A K L V C
- 151 agaattaagg aggattacaa gtgggaattt gaagagcttt tgaaatcctg RIKEDY KWEFEELLKS

- 201 tggtgagetg tttgattett tgaatetggg teaceaggeg etttteeaag C G E L F D S L N L G H Q A L F Q
- 251 agaaggtcat caagactttg gatttttcca caccggggcg cgctgcggct E K V I K T L D F S T P G R A A A
- 301 gctgttgctt ttttgagttt tataaaggat aaatggagcg aagaaaccca A V A F L S F I K D K W S E E T
- 351 tctgagcggg gggtacctgc tggattttct ggccatgcat ctgtggagag H L S G G Y L L D F L A M H L W R
- 401 cggttgtgag acacaagaat cgcctgctac tgttgtcttc cgtccgcccg A V V R H K N R L L L S S V R P
- 451 gcgataatac cgacggagga gcagcagcag cagcaggagg aagccaggcg A I I P T E E Q Q Q Q E E A R
- 501 gcggcggcag gagcagagcc catggaaccc gagagccggc ctggaccctc R R R Q E Q S P W N P R A G L D P
- 551 gggaatga<u>tc taga</u> R E - *XbaI*

(From Figure 15) Nucleotide coding sequence of hamster Bcl2 deletion mutant (SEQ ID NO: 45):

- NCOI
 ccatggctca agctgggaga acagggtatg ataaccgaga gatcgtgatg
 M A Q A G R T G Y D N R E I V M
- 51 aagtacatcc attataagct gtcacagagg ggctacgagt gggatgtggg K Y I H Y K L S Q R G Y E W D V
- 101 agatgtggac gccgcggccg cggccgcgag ccccgtgcca cctgtggtcc G D V D A A A A A A S P V P P V V
- 151 acctgaccct ccgccgggct ggggatgact tctcccgtcg ctaccgtcgc H L T L R R A G D D F S R R Y R R
- 201 gacttcgcgg agatgtccag tcagctgcac ctgacgccct tcaccgcgag D F A E M S S Q L H L T P F T A
- 251 gggacgettt getacggtgg tggaggaact etteagggat ggggtgaact R G R F A T V V E E L F R D G V N
- 301 gggggaggat tgtggccttc tttgagttcg gtggggtcat gtgtgtggag W G R I V A F F E F G G V M C V E
- 351 agcgtcaaca gggagatgtc acccetggtg gacaacatcg ccetgtggat

S V N R E M S P L V D N I A L W

- 401 gaccgagtac ctgaaccggc atctgcacac ctggatccag gataacggag M T E Y L N R H L H T W I Q D N G
- 451 getgggaege atttgtggaa etgtaeggee eeagtgtgag geetetgttt G W D A F V E L Y G P S V R P L F
- 501 gatttetett ggetgtetet gaagaecetg eteageetgg eeetggtegg D F S W L S L K T L L S L A L V
- 551 ggcctgcatc actetgggta cetacetggg ccacaagtga tetaga G A C I T L G T Y L G H K XbaI

Primer 30: CGCAGTACTAGTTTATGGCCTGGGGCGTTTACAGCTC (SEQ ID NO:46)

Primer 31: GAGCTATTCCAGAAGTAGTG (SEQ ID NO:47)